



Team ENERGYneering

Growing Green

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Goal: How can we help solve the world's energy problems??

- Find a way to maximize the production of hydrocarbons
- Find a way to maximize the harvest (yield) of hydrocarbons

Novelty of Our System... Why Algae

E coli

Must be forced to
produce fuels

Algae

Optimized for
the production
of fuel

Yeast

Produces
ethanol
(inferior)



Problem: export is extremely difficult

Novelty of Our System... Possible Solutions

How do we harvest the hydrocarbons?

Option 1:
Lyse the cells
(possibly with a phage gene)

Option 2:
Continuous export
(through metabolic
engineering)

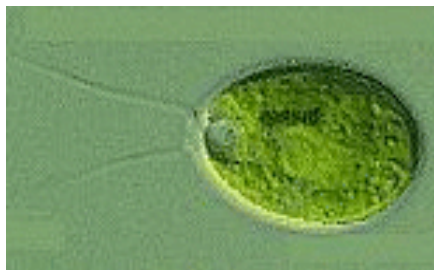


Novelty of Our System... Metabolic engineering

- Goal: force hydrocarbons to be secreted from the algae
- How?
 - Unblock the path by minimizing cell wall
 - Increase the production of hydrocarbons
 - Ramp up hydrocarbon pathway
 - Cut off carbon diversion to other pathways

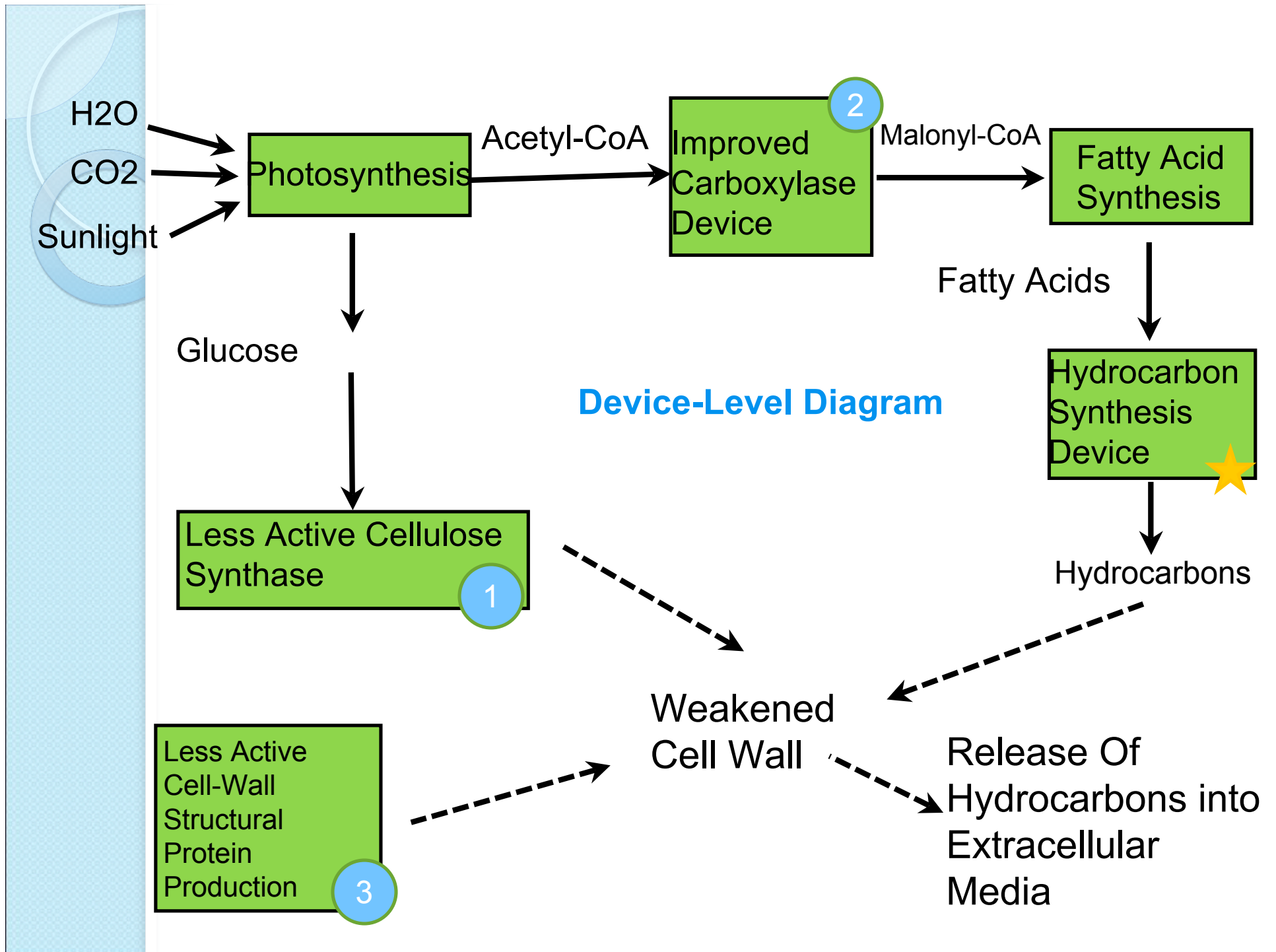
Which species of algae?

- *C. reinhardtii*
- Use as a **model** for our system
- Is already sequenced
- Successful homologous recombination



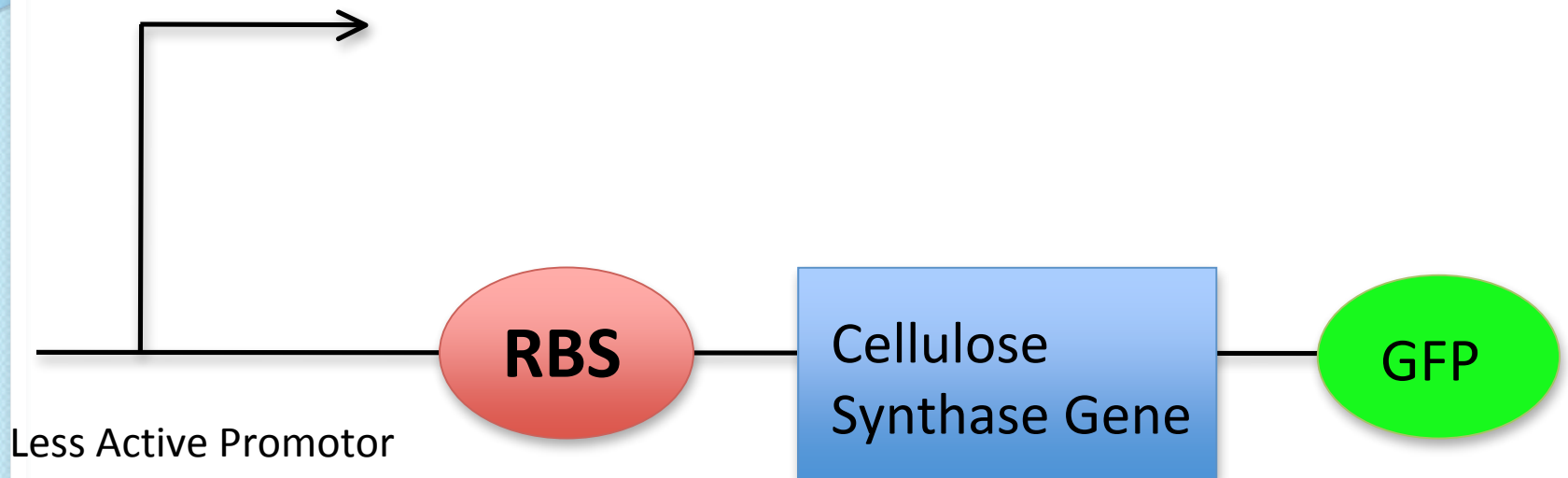
- *B. braunii*
- Our **ideal** species
- Has most productive fuel pathways
- Produces hydrocarbons vs fatty acids
- **Problem**: has not been sequenced





Parts-Level Diagram: Less Active Cellulose Synthase Device

1

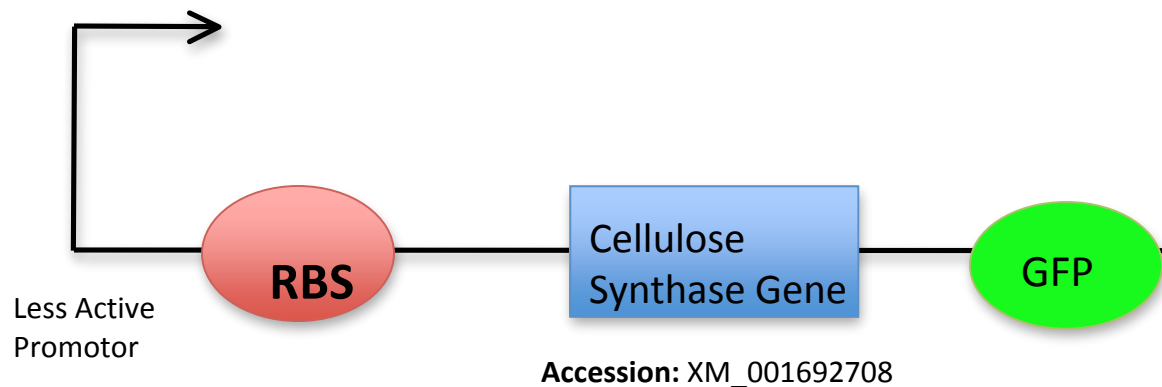


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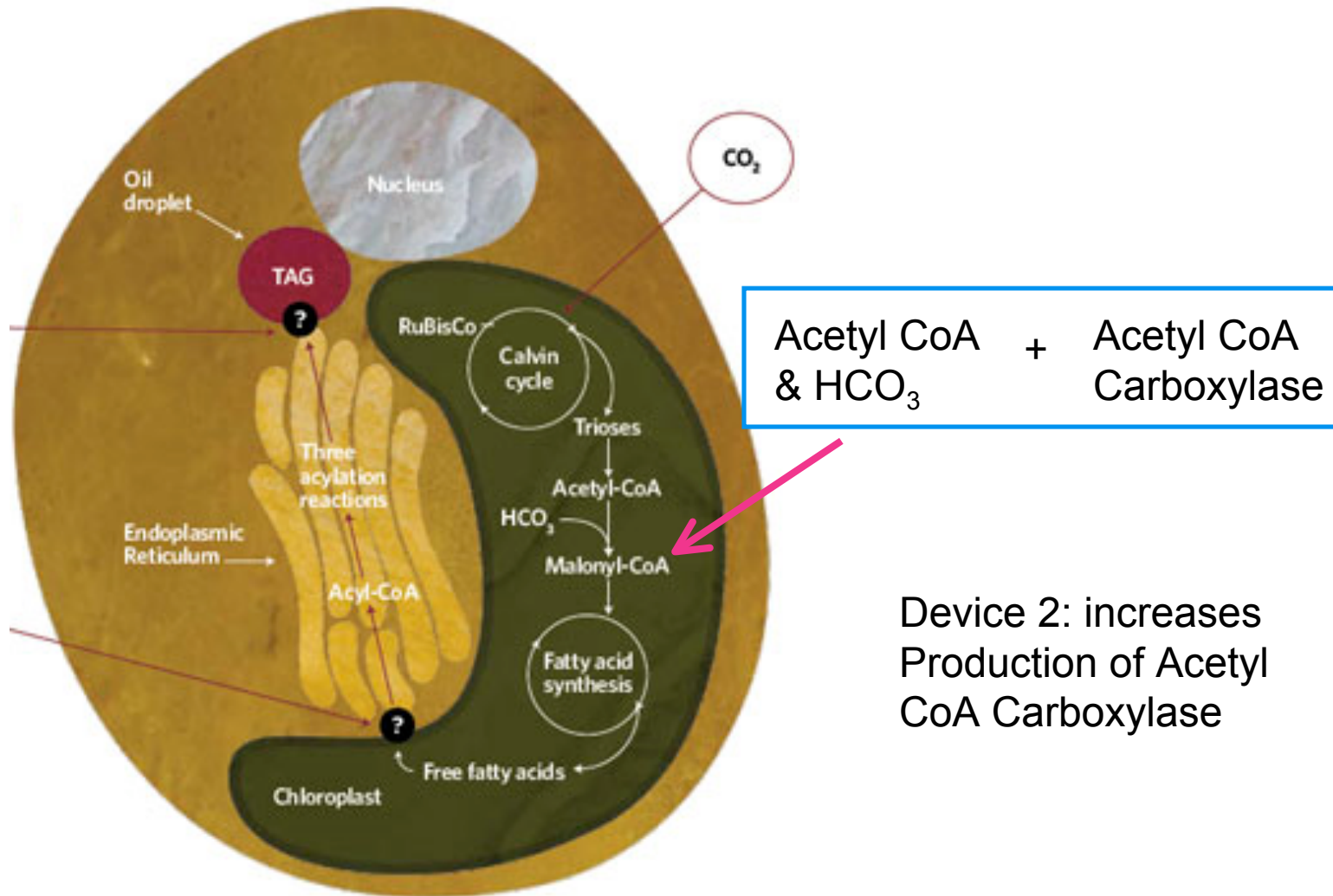
Parts-Level Diagram: Less Active Cellulose Synthase Device

1

- Hydrocarbons are stored in the cell wall
- Decrease cellulose to increase cell wall permeability
 - Test algae promoters (through GFP tagging) to find the least active promoter
 - Ribosome Binding Site: algae specific
- Engineer through homologous recombination

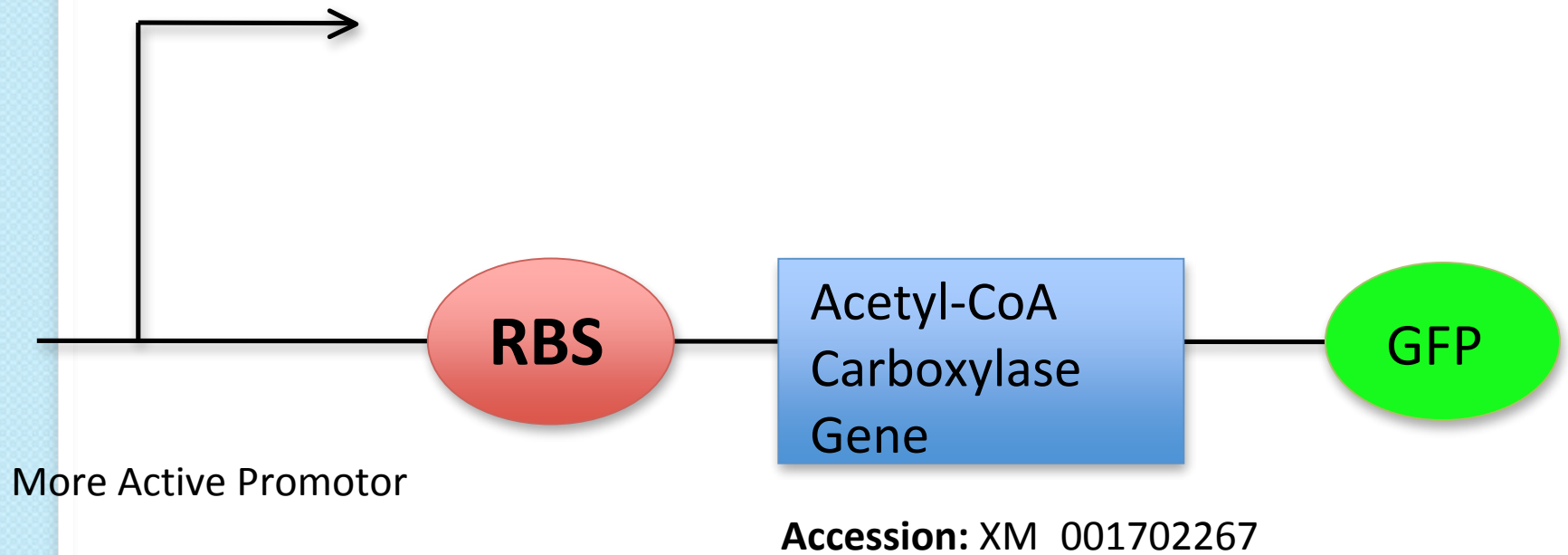


How to increase hydrocarbon production



Device 2: increases
Production of Acetyl
CoA Carboxylase

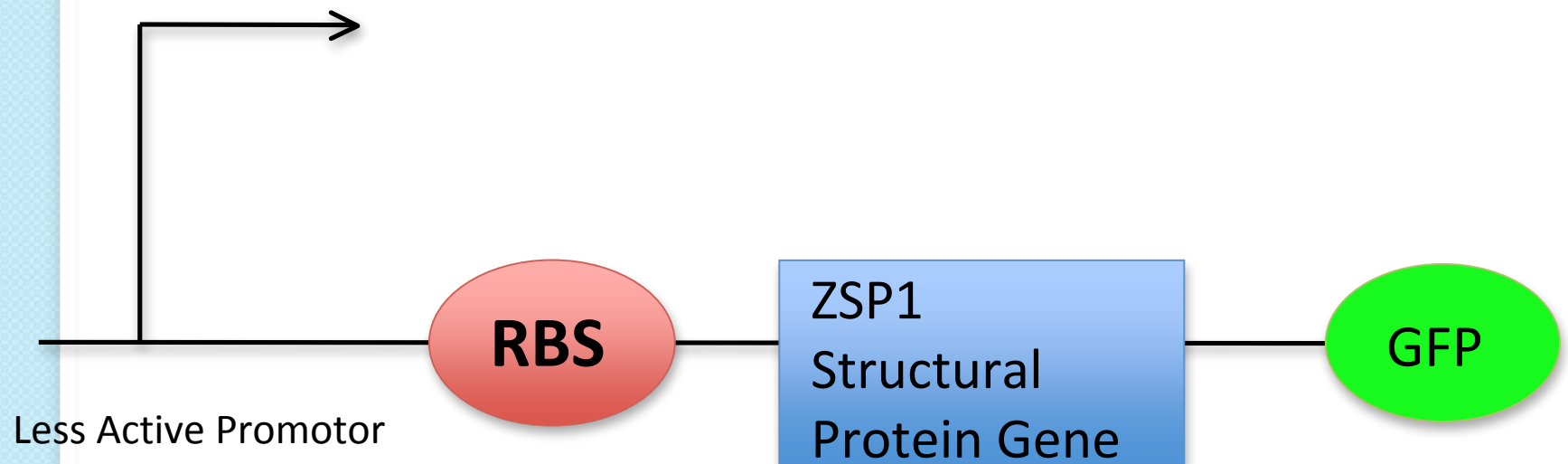
Parts Level Diagram: Improved Carboxylase Device



- Test to find the most active promoter
 - Use GFP to measure the different levels of protein expression
- Incorporate through homologous recombination

Parts Level Diagram: Less Active Structural Protein Production Device

3

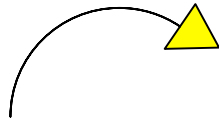


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- Decrease presence of structural protein in the cell wall
- Similarly test for least active promoter and incorporate using homologous recombination

TIMING DIAGRAM

CO₂ Flux

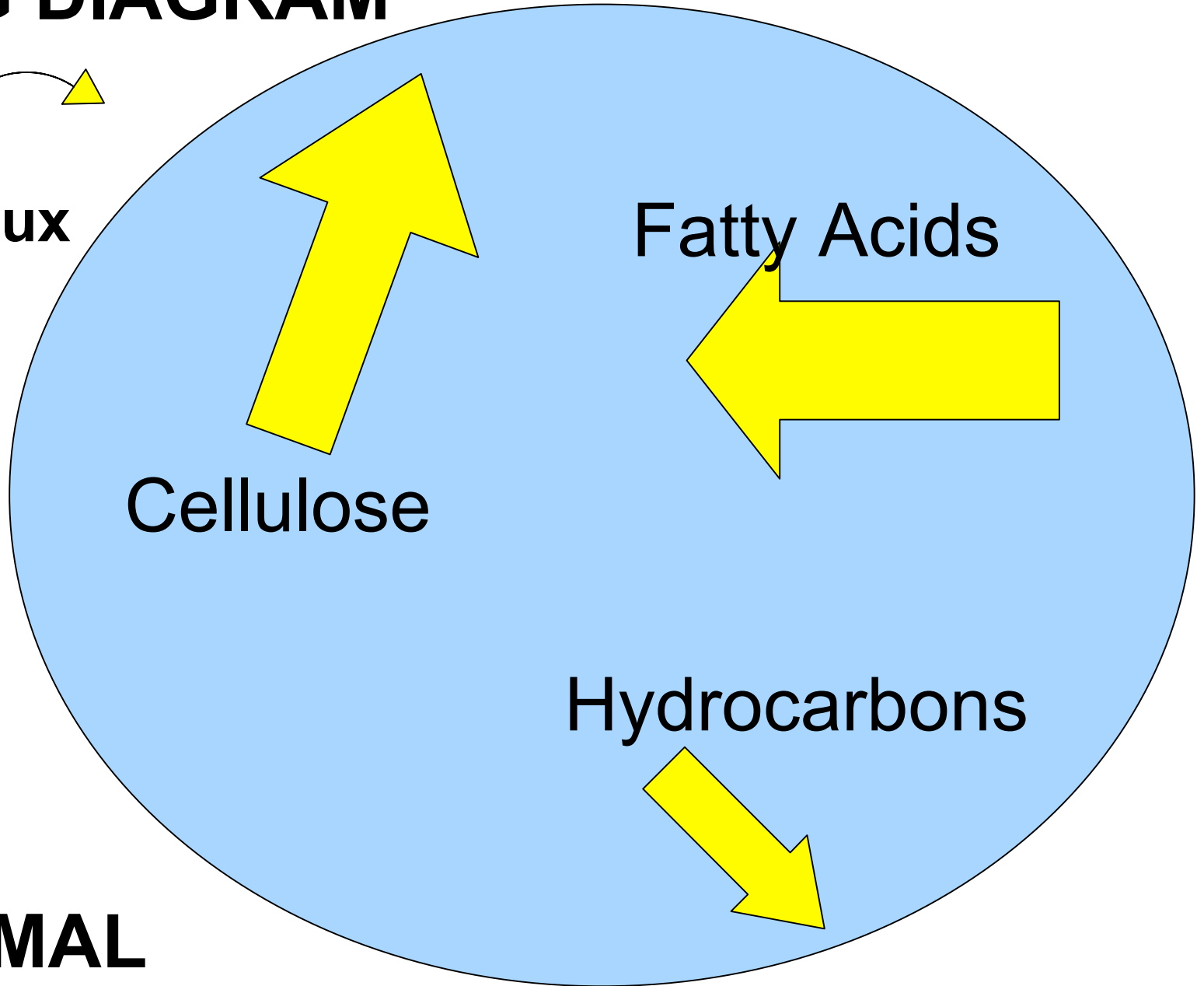


Cellulose

Fatty Acids

Hydrocarbons

NORMAL



TIMING DIAGRAM



Concentration	Change
Cellulose	decrease
Fatty Acids	decrease
Hydrocarbon	increase



Testing and Debugging

- Since we would have established a range of different promoters, we can tune our system to ensure optimum oil production and secretion while maintaining cell's viability
- We can test each device separately to check if they work since they are independent
- Couple the genes of the altered enzymes to different fluorescent genes and test relative expression to see if the device is working according to plan.



Impact of Our System

- Maximize total yield of hydrocarbons
 - Improving rate of production
 - Increasing harvest
- Saving the environment by not using solvents to harvest hydrocarbons
- Decreasing CO₂ emissions



Open Issues

- Ideal species (*B. braunii*) has not yet been sequenced
- Hope to eventually transfer our system into *B. braunii* but we can't be certain that our model will work in both species

Concerns

- **Is it buildable?**

The *C. Reinhardtii* Model is buildable, *B. Braunii* is only buildable after genome sequencing.

- **Time?**

Not a very time intensive project(~3-6 months)

- **Cost:**

Testing the primers could be an expensive process.

However, Overall it shouldn't be too expensive(~15,000\$)

- **Safety:** The algae will be contained in tanks. They should not cause public health issues. Not pathogenic. No new biological material.

- **Security:** Cannot be used to inflict harm. No more of a threat than normal algae.



GO

SOURCES

<http://www.ncbi.nlm.nih.gov/sites/entrez?Db=gene&Cmd=ShowDetailView&TermToSearch=5716257>
http://www.ncbi.nlm.nih.gov/sites/entrez?Db=gene&Cmd=ShowDetailView&TermToSearch=5716257&ordinalpos=4&itool=EntrezSystem2.PEntrez.Gene.Gene_ResultsPanel.Gene_RVDocSum

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<http://www.springerlink.com/content/8x1v7thjvayfgr7p>